BUILDING AN ECONOMICALLY SUSTAINABLE AND INTEGRATED CASSAVA SEED SYSTEM (BASICS-II)

Training Manual on SAH Laboratory Management

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SAHEL Consulting AGRICULTURE & NUTRITION LTD.

BASICS-II

Purpose of Manual

The purpose of this training manual is to serve as a training material for onboarding new cassava processor partners in the BASICS project and a guide for integrated cassava processing companies that are planning to establish and operate a Semi Autotropic Hydroponic (SAH) laboratory for the production of quality cassava stems. It provides a comprehensive overview of the processes and activities involved in the establishment and operation of the SAH laboratory for cassava stem multiplication. It has been developed based on information gathered through research and the insights gained from the operations of a commercial SAH laboratory focused on cassava stem multiplication.

TABLE OF CONTENTS

INTRODUCTION

The processor-led model component of the Building an Economically Sustainable and Integrated Cassava Seed System project (BASICS-II) is focused on establishing an innovative and economically viable commercial stem business model that will serve high quality cassava value chains led by integrated cassava processors.

A rapid propagation technology, known as Semi-Autotrophic Hydroponics, is being implemented to improve the economic viability of stem production and generate processor buy-in by improving supply assurance of high-quality, high starch content cassava roots from its outgrowers and commercial growers.

The integration of a macro-propagation technology into the seed value chain will lower production cost, accelerate the launch of new varieties to markets and encourage the use of other existing improved varieties by smallholder farmers. Also, the use of rapid propagation technology will improve seed production efficiency and enable an economically sustainable cassava seed system.

BASICS-II is the second phase of the four-year BASICS project and IITA is leading implementation of program activities in collaboration with several partners in the public sector and private sector. Sahel Consulting Agriculture and Nutrition Limited is leading the implementation of the processor-led model component. Sahel developed this training manual as a set of curricula on best practices in the management of SAH anchored seed production operations. It is important to mention that these curricula build on the existing body of work developed in the first phase of the BASICS project when Context Global Development led the processor-led model component with support from Sahel Consulting. These best practices curricula are developed around three key areas: business, agronomy, and quality. Business encapsulates the financial aspects of running a financially sound and profitable seed business unit, while agronomy and quality captures the proper multiplication and production practices for quality cassava stems from the SAH laboratory to the field.

The three core curriculum pieces are augmented by a repository of process documentation and specific reports which are developed as tools for helping to standardize operating procedures in accordance with best practices training.

This manual highlights the main activities involved in setting up and operating the SAH laboratory.

Process Flow for SAH Laboratory Setup

In this manual, agronomic related activities within the SAH laboratory are discussed under five main topics. These include SAH laboratory setup, infrastructure, and maintenance; SAH laboratory consumables and materials sourcing; cassava breeder materials sourcing and reception at the laboratory; SAH laboratory operations; and batching and transition of cassava plantlets to the shaded area and nursery. Figure 1 below shows the activity flow diagram for the setup of the SAH laboratory.

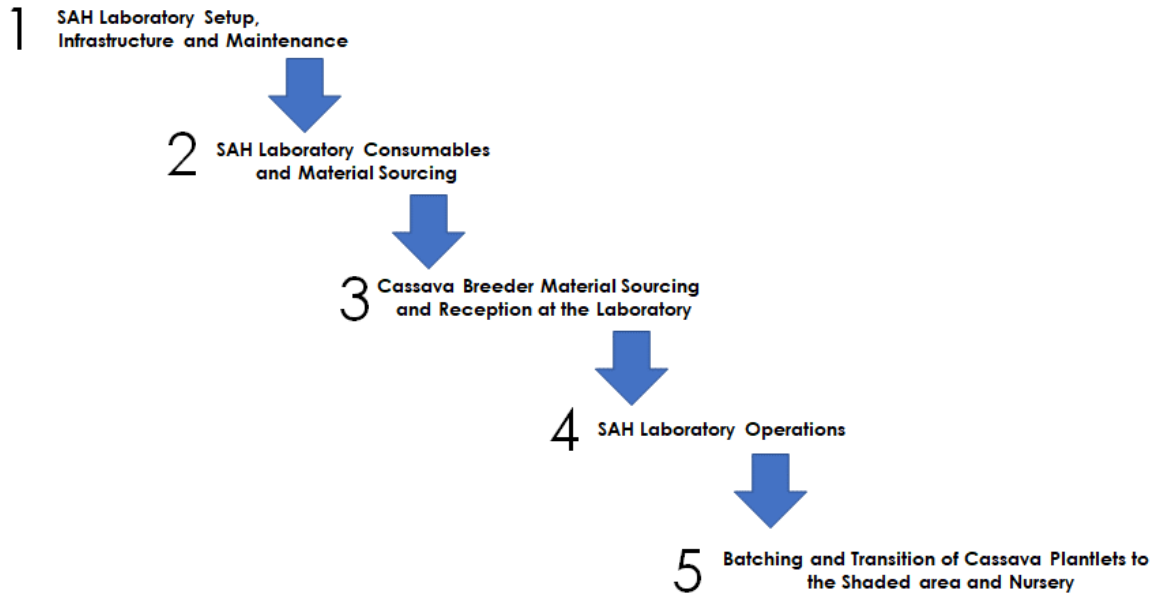

Figure 1. Activity flow diagram for the setup of the SAH laboratory

SAH Laboratory Setup, Infrastructure and Maintenance

Roles

Management, Construction Engineer and Sub-contractors.

Management: The management will be responsible for selecting a suitable site and appointing a construction engineer.

Construction Engineer (Contractor): The construction engineer will be responsible for the development of the SAH laboratory's layout and the construction of the facility.

Sub-contractors: Several sub-contractors will be engaged during the establishment of the SAH laboratory to provide other required infrastructure within the facility. Some of these infrastructures include the rack shelves and alternative power supply system among others.

Activities/Processes

The recommended size of the entire structure housing the SAH laboratory is 374 by 229 inches. It should comprise three main sections:

- The receiving area where mother-plants and laboratory consumables are received.
- The cutting area where plantlets are prepared for multiplication.
- The growth room which houses the racks and shelves where growth boxes are placed.

In addition to the three main sections, a hardening area is important for the gradual introduction of mature plantlets to conditions outside the laboratory for acclimatization. However, with proper training and experience, the hardening area can be eliminated. Other rooms that can be included for operational convenience include a storage room and a convenience room (toilet).

The SAH laboratory setup is presented in the following steps:

1. **Selection of a suitable site**: A suitable site with adequate surrounding aeration should be considered for the establishment of the SAH laboratory facility. The location should have an additional area of 250 by 500 inches size to accommodate the hardening area (temporary shaded area). Establishing the SAH laboratory facility close enough to the nursery field is an important factor to consider when selecting the most suitable site.
2. **Development of a laboratory layout with detailed building specifications**: A detailed layout of the laboratory should be developed to provide clarity on the construction process. The layout should consider all supporting facilities required to run the laboratory including the power generation unit and storage room – for storing inventory of raw materials.
3. **Development of the SAH laboratory building:** A contractor should be engaged to develop the SAH laboratory building. The agreement with the construction contractor should be based on key timelines and milestones to ensure the construction process is completed within a specified timeframe. *Please note that existing buildings and infrastructure such as a metal container can be converted for use as the SAH laboratory.*

4. **Setup of the receiving area:** The receiving area is the first point of reception of all items entering the laboratory. The receiving area will comprise a functional sink for washing hands and rinsing other materials, a locker for placing personal belongings and laboratory footwears for visitors to wear before stepping into the cutting area and growth room.
5. **Setup of the cutting area**: The cutting area is the immediate point of entry to the laboratory from the receiving area. The cutting area is for preparing the growth boxes and substrate, cutting, and multiplying plantlets in old growth boxes into new growth boxes, measuring and mixing chemicals to form nutrient solutions for the plantlets in the growth boxes. It is also a place for storing some raw materials temporary for daily operations in the laboratory. The cutting area is setup with large stainless sheet-surface tables, layers of racks to hold nutrient solutions and chemicals needed for preparing the solutions required in the growth boxes for cultivating the plantlets.
6. **Setup of the growth room**: The recommended size of the growth room is 166 inches square. Layers of racks are installed in the designated growth room to accommodate the growth boxes in layers. The shelves should be designed to ensure ease of reaching boxes in the center of the shelf. The racks are luminated with a special fluorescent bulb fixed across each rack level. The illumination is necessary to provide an artificial light for enabling in the growth of the plantlets. The laboratory is painted in white for adequate illumination. An air conditioning control system should be put in place to control the humidity of the growth room. The growth room should be maintained at room temperature around 26 – 28°C, preferably 28°C.
7. **Setup of the transplanting area**: The transplanting area contains a small space with a flat table desk where growth boxes containing mature plantlets are moved to before transferring to the hardening area (shade area). The transplant area is a biosecurity measure to ensure the gardener handling the transplant process of the mature plantlets to the hardening area does not enter the growth room. In addition, the gardener prepares the necessary growth medium to be used to grow the plantlets in the transplanting area and transfers the nursery trays with the plantlets from the transplanting area to the hardening area.
8. **Setup of the hardening area**: The hardening area is created by erecting a shade with wood or steel reinforcement in form of a flat canopy. The canopy is a thick net material which serves as a membrane to reduce the impact of direct sunlight on the plantlets. As stated earlier, this hardening area can be eliminated if the SAH lab technicians and nursery field workers are adequately trained in transplanting plantlets from the laboratory directly to the nursery.
9. **Development of other supporting infrastructure**: Other supporting facilities are required to ensure that the laboratory is operated optimally. Some of these supporting facilities include:
    - Alternative power supply system;
    - Distilled water system;
    - Internet connectivity.

    Alternative power supply system: The SAH laboratory requires about 13-15hours of electricity supply daily to function optimally. It is important to ensure that an alternative power supply system is put in place to bolster the inconsistent public power supply. Depending on the funds available, a solar power solution might provide a more sustainable solution for stable power supply. However,

diesel/petrol generators are easy-to-install power systems that can be considered. The cost of providing power must be considered when preparing the operation cost forecasts.

Distilled water system: The SAH laboratory requires a continuous supply of clean distilled water for preparing nutrient solutions. A water distiller should be purchased and installed within the cutting area to ensure all-round availability of distilled water.

Internet connectivity: Internet access should be provided through a mobile Wi-Fi device or an independent mobile data connectivity on devices to enable laboratory technicians send periodic data collected from the cutting and batching processes in the SAH laboratory. In a case where the SAH laboratory is situated close to an already existing Wi-Fi coverage owned by the same company, the network can be leveraged to ensure that laboratory technicians are able to send out reports on plantlet cutting and batching every week or any other period based on the preference of the management.

Potential Pitfall and Mitigating Strategies

Pitfall	Mitigating Strategy
Difficulty in sourcing trustworthy contractors especially in the absence of existing relationships with a builder or construction engineer.	• Seek referrals and recommendations from trusted business associates. • Engage the recommended contractors in discussions. • Conduct a due diligence of the shortlisted contractors to confirm their credibility and track record of quality service delivery. • Develop a contract to be signed by both parties.

SAH Laboratory Consumables and Material Sourcing

Roles

Management, Laboratory Material Suppliers, IITA/Research Institutes

Management: The management will be responsible for identifying laboratory material suppliers/vendors and closing on procurement activities. Subsequent contracts can be initialized by the SAH Officer and approved by the management.

Laboratory material suppliers: They are responsible for fulfilling purchasing orders issued by the management.

IITA/research institutes: They are reliable sources of some laboratory materials required for preparing the nutrient solution and substrate for the growth of plantlets in the laboratory.

Activities/Processes

Materials required in the SAH laboratory: These items include laboratory equipment, chemicals, and other consumables. A summary of these items is provided in the table below with corresponding sources where each item can be purchased.

S/N	Item	Description	Source
Equipment (one-time cost)			
1*	Air conditioning system	Helps to ensure the laboratory maintains the required temperature of 26-28°C	General electronic/electrical appliance vendor
2*	Counter-tops with lights	-	Specialized equipment supplier
3*	Adjustable chairs with backrest	For laboratory technicians to sit while carrying out activities in the laboratory like cutting and applying antibacterial disinfectants on the plantlets.	General furniture supplier
4*	Wall thermometer	For monitor the temperature of the growth room	Laboratory material supplier
5*	Timer to control light	To control the lighting of the growth room.	Foreign sourced.
6	UV antibacterial lights or ozone equipment	Not In use	Specialized equipment supplier
7*	Water Distiller	For producing distilled water used to prepare solutions in the laboratory.	
Materials			
8*	Long tweezers	For handling plantlets in the laboratory during cutting.	
9	Round-nosed tweezers	For handling plantlets in the laboratory during cutting.	
10*	Forceps	For handling plantlets in the laboratory during cutting.	
11*	Calibrated plastic beaker	Measuring the solutions for mixing	

12	Plastic graduated cylinder	Not in use	Laboratory material supplier
13*	Small watering can with fine tip	For sprinkling solutions to the mother plantlets	
14	Vapourizer	Not in use	
15*	Disposable syringe	For drawing the acid and the HCl to reduce the pH level and the NaOH to increase the pH level	
16*	Soldering iron wick	To perforate the growth boxes to enhance aeration.	
17*	Portable pH reader or pH measuring tape box	To read the pH level of the solution	
18*	Plastic crates to transfer trays	For carrying growth boxes from the growth room to the cutting area and back, and from the growth room to the transplanting area.	
19	Thermo picnic jug with dispenser	Not in use	
20*	Paper/hand towels	For cleaning and drying the cutting tools after cutting	
21*	Markers	To label the boxes with the appropriate batching information	
22	Glass bottle	Not in use	
23*	Overalls	For protection from contaminations	
24*	Footwears	Used a biosecurity measure to minimize contamination transfer between visitors and the laboratory.	
25*	Plastic containers to prepare nutrient solution	For nutrient solution preparation	
26	Disposable trays with lid (PVC or polypropylene)	Not in use	
27*	Garbage container	For disposing lab waste	
Chemicals (Salts, Micronutrients and Acids)			
28*	Calcium nitrate	To prepare solution A (calcium nitrate + distilled water)	Laboratory material supplier
29*	Potassium nitrate	To prepare solution B.	
30	Monocalcium phosphate	Not in use	
31*	Monopotassium phosphate	To prepare solution B.	
32*	Magnesium sulphate	To prepare solution B.	
33	Copper sulphate	Not in use	
34	Zinc sulphate	Not in use	
35	Boric acid	Not in use	
36	Manganese sulphate	Not in use	
37	Iron sulphate	Not in use	

38	Nutric acid	Not in use	
39*	Hydrochloric acid (HCl)	For preparing solution B	
40*	Ethanol	For cleansing and recycling growth boxes for reuse.	
41*	Antibacterial disinfectant (tween 20)	For disinfecting the plantlets and other contact surfaces like the lab technician hands.	
42*	Fungicide/insecticide/House and Garden spray	To fumigate the growth room.	
Special Consumables from IITA			
43*	Substrate (type; Klassman)	Serves as a component for preparing a rich growth medium for the plantlets	IITA
General Consumables			
44*	Tissue paper	For providing a clean cutting surface while cutting the plantlets in the cutting area of the laboratory.	General consumable merchant/supplier
45*	Hand gloves	Serves as a biosecurity and biosafety measure while handling the growth boxes and the plantlets in the laboratory	
46*	Permanent marker	For labelling the growth boxes to aid traceability in the laboratory.	
47*	Morning Fresh	For sanitization purposes in the laboratory	
48*	Detergent	For sanitization purposes in the laboratory	
49*	Scalpel blades	For handling and cutting the plantlet when producing new growth boxes.	Laboratory material supplier

Table 1. List of Materials and Equipment for the SAH Laboratory

Material Sourcing Procedure:

Following the construction of the SAH laboratory building, the next step is to equip the laboratory with the necessary materials and stock up the initial laboratory consumables required for the first 3-6 months of operations. The volume of laboratory consumables to stock up depends on the storage facility in place to manage inventory. Generally, a storeroom of size 50 by 75 inches will help manage inventory of consumables like substrate for up to three months.

The list provided above is an exhaustive list of all items required to operate a SAH laboratory. The minimum equipment and materials required to operate the laboratory have been highlighted with an asterisk.

Standard Tools and Operating Procedures (STOPs)

Objective: Procedure for sourcing/procuring laboratory materials

1. Develop a list of materials based on the list provided above using the format provided in the table below;

S/N	Name of material	Brief description of material (specification)	Quantity required
1			
2			
3			

2. Create a list of potential laboratory material suppliers and identify potential suppliers through desk research and referrals from key informants such as IITA, Sahel Consulting, Context Global Development, or any operator of a SAH laboratory in Nigeria.
3. Prepare a Request for Quotation (RFQ) to be shared with the potential laboratory material suppliers using the list developed in (1).
4. Send RFQ developed in (3) with the list of potential laboratory material suppliers developed in (2).
5. Follow-up with contact persons in each laboratory material supply company for retrieval of quotations requested based on the RFQ sent.
6. Compare quotations received and select the most appropriate based on criteria such as quality of materials, suitability, price, and terms of transaction etc.
7. Issue a purchase order to the selected supplier(s). The purchase order should specifically define price and specifications of each item, and the terms and conditions guiding the transaction and any additional obligations.
8. Retrieve invoice from supplier(s) and make initial payment instalments based on the agreed terms and conditions of the transaction. Receive receipt for part-payment(s) made to the supplier(s).
9. Expedite the purchase process to ensure adherence with agreed timelines for the delivery of materials. This is done by formally communicating the specific details of the transaction (like payment dates, agreed delivery dates) with the supplier(s).
10. Receive materials from supplier(s) and ensure that items match the specifications provided in the RFQ.
11. Process balance payments and receive receipt for balance payments made.

Potential Pitfalls and Mitigating Strategies

Pitfall	Mitigating Strategy
Unreliable suppliers	• Seek referrals and recommendations from trusted business associates. • Engage the recommended suppliers in discussions and negotiations. • Conduct a due diligence of the shortlisted suppliers by validating suppliers' details and credibility and ascertaining their physical address, years of doing business/operations, obtain feedback and/or referrals from previous clients etc.
Hidden charges	• Validate the terms and conditions of the procurement transaction including the amount of the transaction. • Ensure the supplier signs a binding contract.

Delayed and prolonged transactions	• Request for quotation from the material supply companies and provide deadlines for the submission of quotations. • Follow-up on quotation submission with the sales officer from each of the material supply companies. • Retrieve the quotation and conclude the material sourcing process promptly.

Cassava Breeder Material Sourcing and Reception at the Laboratory.

Roles

Processors (SAH Officer, SAH Laboratory Technician) and Breeder Seed Supplier (Research Institutes).

SAH Officer: The SAH Officer will be responsible for coordinating and supervising the sourcing and transportation of the cassava breeder materials from research institutes. The SAH Officer will also be responsible for coordinating the reception of cassava breeder materials to the SAH laboratory.

SAH Lab Technicians: They will be responsible for the reception of the cassava breeder materials from the research institutes into the SAH laboratory.

Research Institutes: They will be responsible for the production of cassava breeder seed and supply to the SAH laboratory. The manager at the SAH laboratory of the research institutes will be responsible for receiving the request for cassava breeder materials from the SAH laboratory of processors. He will also be responsible for processing and packaging the cassava breeder seed.

Activities/Processes

Cassava Breeder Materials, their Uses and Sources.

Cassava breeder materials for SAH laboratories are sold in two forms - plantlets grown in *in vitro* tubes and SAH growth boxes containing a substrate as shown in figure X below.

Cassava plantlets in *in vitro* tubes

Cassava plantlets in SAH box

Figure 2. Cassava plantlets in in vitro tubes and plastic box

Plantlets in *in vitro* tubes: are produced from the cells or tissues of cassava plants in the tissue culture labs using the tissue culture techniques. *In vitro* cassava plantlets are used as starter materials for the production of SAH cassava plantlets which subsequently produces cassava stems. *In vitro* plantlets grow rapidly, produce exact copies of the mother plants, and are free of diseases and pests. They can be sourced from IITA.

Plantlets in SAH growth boxes: are produced in the SAH laboratory using tissue culture plantlets. They are planted in plastic growth boxes with substrates and transplanted to the field for high quality cassava stem production. The type of substrate used in growing cassava plantlets in growth boxes is known as "Klassman (Sunshine Mix3)". SAH plantlets are more vigorous and

possess the ability to quickly recover from the shock of transplanting to the nursery field. In addition, SAH plantlets have high multiplication and survival rates compared to conventional cassava stem production methods.

Cassava Breeder Material Sourcing Procedure

There are three key activities involved in sourcing for cassava breeder materials for the SAH Laboratory:

- Request for cassava breeder materials
- Transportation of the cassava breeder materials from the source to the SAH laboratory.
- Reception of the cassava breeder materials at the SAH laboratory.

Request for cassava breeder materials: Cassava breeder materials should be sourced from the recommended suppliers. The SAH Officer can either request for *in vitro* plantlets or SAH plantlets depending on the form in which the cassava breeder materials are available at the suppliers' end. Given that SAH plantlets in growth boxes have higher vigour compared to *in vitro* plantlets, processors and other SAH laboratory operators may prefer to request for SAH plantlets in growth boxes to increase the chances of survival of the plantlets in the SAH lab. The request for any of the cassava breeder materials should be made in advance of the total time required for the research institute to process and prepare the order for collection. The SAH Officer should start the request early considering the request procedure of the research institutes, availability, and quantity of the requested variety of cassava at the time of the request and the processing and collection time. The SAH Officer should plan request time to align with the production plan of the lab to ensure continuous operation.

Transportation of the cassava breeder materials to the SAH Lab: The travel distance from the suppliers of the cassava breeder materials to the SAH lab and the prevailing environmental conditions during transit are major factors to be considered during transportation. The SAH lab technician should use a properly sanitized vehicle to transport the cassava breeder materials from the source to the SAH lab. Cooling vans are preferred for transporting the cassava breeder materials. In the absence of cooling vans, the cassava breeder materials should either be in the growth boxes or *in vitro* tubes, sealed and packed in a cooler before loading into the vehicle. Carton boxes could also be used to pack SAH growth boxes or *in vitro* tubes in the absence of cooling systems. Proper care should be taken when loading the cooling systems or cartoons containing the cassava breeder materials into the vehicle. The SAH lab technician should verify the accuracy of the order by confirming that the requested cassava varieties and order quantity are processed before leaving the premises of suppliers. Transportation of the cassava breeder materials should be done during the cool hours of the day to minimize the effects of harsh environmental conditions on the plantlets.

Reception of the cassava breeder materials at the SAH Lab of the processors: The receiving area of the SAH lab should be thoroughly cleaned and disinfected a week before the arrival of the new cassava breeder materials to prevent contamination. On the arrival of the cassava breeder materials to the SAH Lab, the packing carton boxes, or coolers should be offloaded into the receiving area. The SAH growth boxes or *in vitro* tubes should be unpacked and transferred to the growth room. The plantlets should remain in the growth room for 2-3 days before cutting commences.

Standard Tools and Operating Procedures (STOPs)

Objective 1: Request for cassava breeder materials

The following steps should be taken when requesting for cassava breeder materials:

1. Contact the lab manager at the research institute to make a request.
2. Send an email to the lab manager providing the following details for the request;
    a. Name of requestor
    b. Name of the requesting SAH Lab
    c. Purpose of use
    d. Name of varieties and quantity of each variety.
3. Follow-up on the request email sent to the lab manager at the research institute to confirm the available quantities of the cassava varieties and types of breeder materials.
4. Read and sign the terms and conditions form.
5. Place the order and make the first tranche of payment.
6. Obtain confirmation for the payment and date for the collection of the order.
7. Follow-up the order until the date of collection.
8. On the day of collection, check the order and ensure that the quality control procedures below are in place. Confirm that;
    a. the requested cassava breeder materials are in good condition.
    b. the specified varieties and quantity of cassava breeder materials are well packed in sealed cartoons or cooling systems with the careful handling signs 'UP ARROW' and 'FRAGILE' written on each pack[1].
    c. one variety of cassava is contained per box and each box is labelled with the following information: name of the variety, number of SAH boxes or *in Vitro* tubes and date of production and have the phytosanitary certificate of the cassava breeder materials.
9. Ensure that the phytosanitary certificate of the cassava breeder materials is provided by the supplier.
10. Receive the order.

Objective 2: Transportation of the cassava breeder materials to the SAH Laboratory.

The following steps should be taken when transporting the cassava breeder materials to the SAH labs of the processors:

1. Carry out both mechanical and hygiene check on the vehicle that will be used to transport the cassava breeder materials prior to collection.
2. Prior to loading, the SAH lab technician should carry out quality control procedures to ensure the accuracy of the order.
3. The packing boxes should be arranged in an upright position inside the vehicle and should be kept from direct sunlight[2].
4. For long distance transportation, lasting more than 2 days, the packing boxes should not be completely sealed. The boxes should be kept slightly open and the windows of the vehicle also opened to allow air ventilation and exposure to sunlight. This is because

[1] Postflask Management of Cassava and Yams, IITA, 2002
[2] Postflask Management of Cassava and Yams, IITA, 2002

long periods of darkness may result in loss of vigour for the cassava plantlets and later reduce survival rate particularly for plantlets *in vitro* tubes[3].

Objective 3: Reception of the cassava breeder materials at the SAH Lab of the processors

The following steps should be taken when receiving the cassava breeder materials into the SAH lab:

1. Prepare the receiving area before the arrival of the cassava breeder materials.
2. Offload the packing boxes into the receiving area.
3. Sanitize the packing boxes with mist spray of alcohol before unpacking the growth boxes or *in vitro* tubes[4]
4. Keep a record of the received breeder materials into the lab logbook with the following information; the name of cassava variety, number of each variety, date of production, the number on the SAH boxes or *In vitro* tubes and date of reception into the lab.
5. Transfer the SAH boxes or *in vitro* tubes to the growth room and place them under fluorescent light at a low relative humidity and temperature between 22^0C-28^0C to allow the cassava plantlets acclimatize to the new environment and recover from transportation stress of before cutting.

Potential Pitfalls and Mitigating Strategies

Pitfall	Mitigating Strategy
Delay in the collection of order due to the unavailability of desired cassava varieties and volume required at the time of the request.	• Forecast and plan for the required varieties and volumes of breeder cassava varieties yearly. • Ensure the request process for breeder cassava varieties are initialized ahead of the time of need.
Cross-contamination of varieties due to poor handling during transportation	• Ensure adequate packaging and handling during transportation of cassava breeder material from the research institute to the SAH laboratory. • Ensure biosecurity measures are taken in the process of transporting and transferring cassava breeder materials from the research institute to the SAH laboratory.

[3] Micropropagation and Hardening Sweetpotato Tissue Culture Plantlets; S. Namanda, R. Gatimu, S. Agili, S. Khisa, I. Ndyetabula, and C. Bagambisa; 2015
[4] Micropropagation and Hardening of Sweetpotato Tissue Culture Plantlets.

Laboratory Operations

Roles

SAH Laboratory Technicians, SAH Laboratory Officer and Gardener.

SAH Laboratory technicians: They will be responsible for carrying out daily activities which include;

- cutting of plantlets and cutting plantlets into new growth boxes,
- monitoring of plantlets in the growth room,
- removal of withering plantlets and application of antibacterial disinfectants on plantlets in the laboratory.

Technicians are also responsible for managing the transfer process of plantlets from the growth room to the transplant area.

SAH Officer: The SAH Officer will be responsible for managing and overseeing activities in the SAH laboratory, the hardening area and the SAH nursery.

Gardener: The Gardener will be responsible for preparing the growth medium in the nursery trays ahead of the transfer of plantlets from the transplant area of the lab to the hardening area outside the lab. The gardener also manages activities such as watering of plantlets in the hardening area.

Activities/Processes

Plantlet routine management

After receiving the cassava breeder seed from the supplier, the plantlets are multiplied in the laboratory through a series of activities. These activities include the following;

Preparation of solutions to be applied to the substrate: In preparing a stock solution to serve as nutrients to the substrate (the growth medium for the SAH plantlets), solution A (500ml), solution B (500ml) and ordinary water (1,000ml) are mixed together. The resulting solution is used to treat the substrate which forms the recommended growth medium for the plantlets.

Cutting of the plantlets and re-boxing: The stepwise approach adopted in multiplying the SAH plantlets in the laboratory is provided below;

1. Spread tissue towels on the table and spray with Tween 20 to dampen and disinfect the tissue ahead of cutting activities.
2. Move the mature plantlets from the growth room to the cutting room.
3. Micro-ratoon the mature plantlets into two parts. Cut the upper part detached from the growth medium into 2 parts; the apical and auxiliary part.
4. After cutting, plant the apical and auxiliary parts separately into new growth boxes because the apical part grows faster than auxiliary part. If planted together, their growth will not be uniform.
5. After transplanting, spray the newly transplanted plantlets with Tween 20 and close the lid of the growth box tightly.
6. Add 100ml of nutrients to the mother plant after cutting from it and spray with Tween 20.
7. Take both the mother box and the new box back to the growth room.

Recording of data on plantlets and growth boxes: As new growth boxes are created, the boxes are labelled and stored in a specific batching pattern across the shelves in the growth room. In addition, data on the number of new growth boxes created and the relationship with the mother plants is entered into a software[5] used for collecting and recording data on the SAH plantlets in the laboratory.

Monitoring of plantlets' growth in the SAH Lab's growth room: Laboratory technicians are required to carry out occasional monitoring rounds in the growth room to observe the performance of the plantlets. Some of the key signs the laboratory technicians should look out for and possible remedies include;

Conditions	Solutions
Plants etiolating[6]	Add nutrient solution or water to the substrate
Plantlets wilting and drying up	Add water to the substrate
Plantlets yellowing	Add nutrient solution to the substrate
Temperature and humidity in the growth room	If humidity is higher than required level in the growth room, increase the air conditioning temperature output
Dry substrate	Add nutrient solution[7]

Table 2. Monitoring of plantlets in the SAH growth room

Biosecurity management: Biosafety and biosecurity measures are required to manage and mitigate disease outbreak and other biological contamination that can possibly result from exposing humans to possible contamination in the laboratory while ensuring that contamination from humans as host is prevented through some set measures to guide operations within the laboratory. Considering the possible transfer of contamination that can occur through the introduction of new subjects (visitors/unfamiliar humans) into the laboratory, visitors should be properly screened before allowed access into the laboratory. The screening process will involve disinfecting their hands with a solution mixture of Tween 20 and detergent. In addition, a cleansing procedure involving the application of the T ween 20 and detergent solution should be applied to the hands of any visitor who intends to move from one growth room to the other.

Standard Tools and Operating Procedures (STOPs)

Objective 1: Receiving visitors from outside the SAH laboratory.

1. Visitors[8] are accepted through the receiving area.
2. Visitors' footwear should be replaced with a laboratory shoe before leaving the receiving area into the SAH cutting area and into the growth room.

[5] FieldBook is an open-source software application used for taking records of the plantlets in the SAH laboratory.
[6] Plantlets having loss of vigor or pale appearance.
[7] Solution A + Solution B + ordinary water
[8] A visitor is anyone who does not work in the SAH laboratory and seeks to access the SAH laboratory with granted permission.

3. Visitors are required to wash their hands using detergent and rinsing with ordinary water in the basin located in the receiving area and /disinfect their hands with alcohol mist.
4. Visitors are required to put on lab coat before entering the cutting area or the growth room.

Objective 2: Movement from one growth room to another (in case of multiple growth rooms)

1. Laboratory technicians/visitors/any other personnel should sanitize their hands by applying available antibacterial disinfectant to their hands before moving from one growth room to another.
2. ALL PERSONS in the growth room at any point in time should put on disposable hand gloves before touching any plantlet or growth box in the growth room.

Environmental management: Keeping the vicinity of the laboratory clean is critical to the biosafety of the SAH laboratory. The growth room is cleaned every two days using Tween 20 solution and a small quantity of detergent. The waste bin is placed outside the laboratory building to hold laboratory waste. The waste bin should be emptied every two days or on an as-required basis.

Standard Tools and Operating Procedures (STOPs)

Objective: Cleaning the growth room floor

1. Prepare the cleaning solution to mop the growth room floor by mixing 5ml of detergent with 10ml of Tween 20.
2. Apply the cleaning solution to the mop and mildly scrub the floor of the growth room (The cleaning solution will be sufficient for mopping a 20-sqm floor).
3. Allow the floor 3-5 minutes to dry up before granting access to anyone into the growth room.
4. Mop the growth room's floor every two days.

Potential Pitfalls and Mitigating Strategies

Pitfall	Mitigating Strategy
Granting access to unauthorized visitors into the laboratory.	• Provide full lab coats to all visitors before granting access into the laboratory. • Train the security staff and ensure that they are aware of the protocol to access the facilities.
Keeping waste items in temporary nylons within the laboratory.	• All waste should be disposed in the waste bin outside the laboratory and emptied every two days as recommended.

Batching and Transition to Shaded Area and Nursery

Roles

SAH Lab oratory Officer, Laboratory Technicians, Gardener, Nursery Supervisor, Nursery Field-workers.

SAH Officer: The SAH Officer will be responsible for coordinating and supervising the batching and transfer of the plantlets from the plantlets from the SAH laboratory to the hardening area and to the nursery field. The SAH Officer will also be responsible for coordinating the management of the plantlets in the hardening area and supervising all operations in the nursery.

SAH Laboratory Technicians: They will be responsible for batching the plantlets in the lab and moving the plantlets in the growth boxes from the lab to the transplanting area.

Gardeners: They will be responsible for preparing the nursery tray and transplanting of the plantlets from the growth boxes into the nursery trays. He will also be responsible for the daily monitoring and management of the plantlets in the hardening area.

Nursery Supervisor: The Nursery Supervisor will be responsible for receiving the plantlets at the nursery field and the management of the field workers maintaining the nursery field.

Nursery Field Workers: They will be responsible for transplanting the plantlets from the nursery trays to the nursery field and routine maintenance of the plantlets on the nursery field.

Activities/Processes

Plantlet Batching Procedure in the SAH Laboratory, the Hardening Area, and the Nursery

This is an important activity in the propagation and multiplication cassava plantlets to produce high-quality stem. It is a way to enhance proper identification and management of the plantlets from the SAH laboratory to the hardening area and to the nursery field. The plantlets are batched based on the information written on the growth boxes. This information includes; name of variety, growth box number, planting date, substrate number and initials of the name of laboratory technician that planted into the growth box. This information changes after each cutting of the plantlets, and as they are transplanted to the hardening area and to the nursery field.

Standard Tools and Operating Procedures (STOPs)

Objective 1: Labelling the growth boxes in the SAH Laboratory.

1. On the reception of the cassava breeder materials, enter the following information; the name of the variety, number of varieties, date of production, the source of the breeder materials, the number of the growth box and date of reception into the lab written on each of the growth boxes into the software boxes and proper recording of the data on the boxes into the data collection software application[9].

[9] The software currently used is Field Book downloadable on Google PlayStore.

2. After cutting the plantlets in each growth box, label the new growth box with the following information; the name of variety, growth box number, planting date, substrate number and initials of the of the lab technician and enter them immediately into the software application.

Objective 2: Labelling the nursery tray in the Hardening area.

1. Assign a number to each nursery tray.
2. Label each nursery tray with the name of the variety, source of variety, date of transplanting, number of the nursery tray, and the number of the growth box from which the plantlets are transferred.

Objective 3: Labelling the planting plots on the Nursery field.

1. Partition the nursery field into smaller planting plots and assign a number to each of these plots. The assigned number should be serially assigned (e.g., 201, 202, 203, 204 etc.)
2. Place a signpost on each plot with the following information: name of the variety, the source of variety, date of planting, number of nursery trays and plot number.

Plantlet Transfer Procedure from the SAH Laboratory to the Hardening Area

The hardening area is where the plantlets are acclimatized to the natural environment before transferring to the nursery field for field stem production and multiplication. The hardening process involves adapting the plantlets to the natural environment by gradually minimizing the supply of water and nutrient, decreasing temperature, and increasing light intensity. The plantlets are moved to the hardening area 2-3 weeks (depending on the variety) after their third cutting. The hardening area is a shade covered with a green net right outside the SAH laboratory. It is advisable to use green net for the shaded area because it enhances the adaptation and adjustment process of the plantlets to the field environment. Please note that the plantlets are already pre-hardened through the SAH procedure in the laboratory, hence it is recommended that the plantlets are kept in the hardening area for 2 weeks before transplanting to the nursery field.

Standard Tools and Operating Procedures (STOPs)

Objective: Transplanting the plantlets from the SAH lab to the hardening area.

1. Put on protective gloves sterilized with alcohol spirit to avoid any possible contamination of the plantlets.
2. Take stock and sort out the plantlets in the growth room that have been cut thrice (3 times).
3. Move the sorted plantlets to the transplanting area.
4. Check the nursery trays to ensure they are professionally cleaned and disinfected against any possible disease infection from previous use.
5. Fill the nursery trays with sterile and free draining compost soil mix then moisten with water. This compost soil mix can be made from the mix any of these components; sand, cocopeat, perlite, vermi-compost, and vermiculites. For cassava, a porous mix of 3 parts of perlite/vermiculite or sand and 1-part potting mix or soil[10].
6. Carefully remove the plantlets from the growth boxes with the use of a forceps or tweezer to avoid damaging the roots.

[10] The Transfer of Tissue Culture from Tube to the Soil.

7. Check the length of the plantlets' roots and trim the ones with very long roots. This would help prevent the roots from damaging during planting, prevent the plantlets from dying and allow the formation of new roots once the plantlets are established.
8. Pre-treat the plantlets by lightly dipping their root base into a fungicide to prevent any possible fungal infection.
9. Make a hole with your thumb and place the roots into the soil-filled nursery trays. Gently and carefully firm-up the soil around the roots by pressing the surface of the soil with the thumb.
10. Ensure each nursery tray is containing the same variety of cassava plantlets and they are properly labelled using a waterproof label inserted in the nursery tray with the following information: name of variety, source of variety, date of transplanting, number of the nursery tray and the number of the growth box from which the plantlets are transplanted.
11. Cover the shaded area with green net for the next 2 weeks to maintain high humidity and help the plantlets to adapt to the new environment.
12. Perform daily routine management practice (watering and disinfecting).

Management of Plantlets in the Hardening Area

This involves the daily routine agronomic practices carried out to ensure the effectiveness and the optimal performance of the plantlets in the hardening area. These agronomic practices include watering of the plantlets, removal of outliers and dry/dying plantlets and application of control measures against diseases and pests.

Standard Tools and Operating Procedures (STOPs)

Objective: Managing the plantlets in the hardening area.

1. Water the plantlets daily (preferably in the morning and evening) to prevent dryness of the growth mediums.
2. Inspect the plantlets daily to ensure the net used in the shaded area is in proper shape.
3. Observe the plantlets at regular intervals (morning and evening) to check for any signs or symptoms of nutrient deficiency or disease infection and take appropriate action when any symptom is noticed (removal of dying plantlet or watering).
4. Keep daily records of all observation and agronomic practices carried out in the field book.

Plantlet Transfer Procedure from The Hardening Area to the Nursery Field

The plantlets are transferred to the nursery field after the 2 weeks of hardening in the shaded area. The nursery field represents the first full exposure of the plantlets to the external natural growth environment for stem production. The distance between the SAH laboratory and the nursery field should be considered when designing the SAH system. This distance determines the mode of transferring plantlets from the hardening area to the nursery field. It is advisable that the hardening area is in good proximity to the nursery field to minimize the stress on the plantlets due to distant travel during transplanting. The plantlets should be transported to the nursery field with a pick-up vehicle (in a case where the distance from the hardening area to the nursery field is more than 100m, otherwise, the plantlets can be carried by hand for distances under 100m). It is recommended that the transporting and transplanting of the plantlets to the nursery field should be done early in the morning before sunrise or in the evening immediately after sunset (preferably for distance under 100m only). The site for the

nursery field should be well-drained, weed-free and located near reliable source of water for irrigation ahead of the dry season.

Standard Tools and Operating Procedures (STOPs)

Objective: Transferring plantlets from the hardening area to the nursery field

1. Clean the pick-up vehicle to ensure it is free from any possible contaminants. Spray with an antibacterial disinfectant to eliminate possible infections.
2. Gently load the plantlets in the nursery trays into the vehicle and ensure the waterproof labels on the nursery tray do not falloff in the process.
3. Drive the vehicle carefully to the nursery field.
4. Divide the nursery field into plots to allow for the transplanting of one variety per plot.
5. Label each of the plot with a signpost having the following information; name of variety, source of variety, date of planting and number of nursery tray.
6. Off-load each of the nursery tray to the designated plot where the plantlets will be transplanted.
7. Carefully remove each plantlet from the cells of the nursery tray with some ball of soil around its root and transplant.
8. Water the plantlets immediately after transplanting.

*Note that #4 & 5 should occur prior to arrival at the nursery field. This activity will be carried out by the nursery supervisor.

Potential Pitfalls and Mitigating Strategies

Pitfall	Mitigating Strategy
Varietal mix-up when batching the plantlets in the SAH laboratory, hardening area and nursery field.	• Batching should be done separately for each variety. Simultaneous batching of multiple varieties should be avoided at all times.
Stress resulting from the transfer of the plantlets from the hardening area to the nursery. *(In cases where the distance between the hardening area and the nursery is more than 100m)*	• Ensure the vehicle is properly aerated to minimize stress on the plantlets when transporting the plantlets from the hardening area to the nursery field
Stress resulting from placing the plantlets in an unconducive area upon arrival at the nursery field.	• Ensure the plantlets are transferred from the vehicle to a cool shaded area around the nursery field in cases where the plantlets will not be transplanted to the nursery plots immediately on arrival.

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